

Modeling PRRSV-2 prevalence within a farrowing room

Onyekachukwu H. Osemeke¹, Eduardo de Freitas Costa², Vinicius Weide³, Swaminathan Jayaraman¹,
Gustavo S. Silva¹, Daniel C. L. Linhares¹

¹Veterinary Diagnostic and Production Animal Medicine, College of Veterinary Medicine, Iowa State University, Ames, Iowa, USA.

²Department of Epidemiology, Bioinformatics and Animal Models, Wageningen Bio-veterinary Research, Lelystad, The Netherlands.

³Instituto Federal Rio Grande do Sul

ABSTRACT

Estimated disease prevalence is an important assumption in calculating the minimum number of animals/units to be sampled for PRRSV surveillance/monitoring programs (Christopher-Hennings et al., 2001; Holtkamp et al., 2011, 2021; Rotolo et al., 2017; Rovira et al., 2007); these programs consider the minimum proportion of animals within a herd that is likely positive (viremic) for PRRSV to estimate the number of animals to be sampled. Family oral fluids (FOFs) have been demonstrated to be a more practical sample type for low-prevalence PRRSV surveillance in weaning-age pigs (Almeida, Zhang, Zimmerman, et al., 2021; Osemeke et al., 2022), since this sample is an aggregate sample type collected at the level of the crate/litter, the minimum number of litters/units to be sampled (sample size) will depend on the estimated number of positive litters/crates in a farrowing room, the diagnostic performance of the test, and a chosen confidence level (Osemeke et al., 2022; Rotolo et al., 2017).

This study aimed to characterize a relationship between the proportion of PRRSV-positive (viremic) pigs, the proportion of PRRSV-positive litters, and the likely proportion of litters to be positive by a FOF RT-rtPCR test in a farrowing room, using Monte Carlo simulations and data from an earlier study (Almeida, Zhang, Lopez, et al., 2021). Expectedly, the spatial distribution (clustering parameter) of viremic pigs within a farrowing room influenced the proportion of litters having at least one viremic piglet, and the number of positive FOFs expected from the room. This study assessed and measured this clustering parameter from the reference study and applied

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30 different values of the clustering parameter to the stochastic models to estimate a relationship
31 between all three proportions.

32 The results of this study provide guidelines on how practitioners can ascertain the number of FOF
33 samples needed to match serum samples at an assumed piglet-level prevalence and clustering level.
34 The results from this study also provide a platform for swine practitioners to estimate the likely
35 proportion of viremic pigs in their barns, given the PRRSV-RT-rtPCR positivity rate of FOF
36 samples submitted from a farrowing room.

37 From our findings, an estimated level of clustering of viremic pigs is worth considering when
38 estimating the litter-level prevalence of PRRSV for onward FOF sample size calculations.

39

40 **Introduction**

41 Porcine respiratory and reproductive syndrome virus (PRRSV) poses a significant challenge to the
42 global swine industry (Calderón Díaz et al., 2020; Holtkamp et al., 2013). Monitoring/surveillance
43 remains an integral component of PRRSV control and elimination programs, and ascertaining the PRRSV
44 status of pig populations around the time of weaning is crucial to guide decisions on health
45 interventions and pig flow (Holtkamp et al., 2011).

46

47 Efficient PRRSV surveillance/monitoring programs allow for the early detection of infection
48 and helps evaluate changes in PRRSV prevalence over time; aiding swine producers and
49 veterinarians alike to forestall the spread of PRRSV (Mccaw, 2000; Silva et al., 2017), and evaluate
50 progress made with instituted PRRSV management programs (Holtkamp et al., 2021; Linhares et
51 al., 2014).

52

53 Different sample types are routinely submitted to Veterinary Diagnostic Laboratories in the US for
54 PRRSV investigation by RT-rtPCR; these would include samples taken from individual pigs such
55 as serum, swabs, semen, and post-mortem tissues; or aggregate samples taken from multiple pigs
56 such as processing fluids and oral fluids (Trevisan et al., 2019). These samples are either submitted
57 and tested individually or in pools.

58

59 The number of samples submitted for disease pathogen investigation is crucial to the success of a
60 surveillance/monitoring exercise. Guided by epidemiological/statistical assumptions, the sample
61 size should have enough power to detect at least one positive unit if the herd is truly positive for
62 the pathogen of interest (Cameron et al., 2020; Stevenson, 2021).

63 Estimated prevalence at the individual pig level is one of the key variables used in calculating
64 sample size to demonstrate disease freedom (Fosgate, 2009; Stevenson, 2021).

65 The preferred sample for PRRSV surveillance in sow herds is serum from weaning-age pigs
66 (Holtkamp et al., 2011). Although the serum sample is the sample of choice for PRRSV
67 surveillance, it requires more skill, more manpower, is less animal welfare friendly, and is often
68 impractical for frequent PRRSV surveillance in large herds (Turlewicz-Podbielska et al., 2020)
69 compared to other (alternative) sampling options. For these reasons, since 2018, validated
70 aggregate alternative samples have been the most frequently submitted samples for PRRSV
71 surveillance in the US (Trevisan et al., 2019).

72

73 ~~Almeida~~ (Nunes de Almeida, (2020) demonstrated that, especially in low prevalence, ~~Family~~
74 family oral fluids (FOFs) are a more convenient and cost-efficient alternative to serum sampling
75 for PRRSV surveillance in weaning-age pigs. A FOF sample is an aggregate sample obtained when
76 oral fluids are wrung off a rope chewed by a sow and her piglets (Almeida et al., 2020). A challenge
77 with interpreting a positive result from testing FOF and other aggregate sample types is that one
78 only knows that at least one animal that contributed to the sample matrix is pathogen-positive
79 (shedding) but cannot ascertain the exact number of animals shedding.

80 Consequently, little to nothing is known about the number of positive pigs in a sampled room, given
81 the proportion of PRRSV-positive aggregate samples, such as FOF, obtained from that room.

82

83 The individual pig is the unit for which sample size is calculated when non-aggregate samples are
84 collected, while the litter is the unit for which sample size is calculated when an aggregate sample
85 such as FOF is to be collected (Osemeke et al., 2022; Rotolo et al., 2017) it will be helpful to swine
86 practitioners in making sampling decisions if they understood how the proportion of PRRSV-
87 positive (viremic) piglets related with the proportion of PRRSV-positive litters, as both parameters
88 are needed assumptions in estimating sample sizes.

89
 90 To the best of our knowledge, the relationship between the mentioned proportions in a swine
 91 farrowing room has not been previously characterized. This study builds upon a previous study that
 92 assessed the observed probability of a positive FOF sample given the number of viremic PRRSV
 93 RT-rtPCR positive piglets within a litter (Almeida, Zhang, Lopez, et al., 2021). We further
 94 developed an *in-silico* study to assess and model a relationship between the Piglet-piglet level
 95 prevalence (PP), True-true litter-level PRRSV prevalence (TLP) and Apparent-apparent litter-level
 96 PRRSV prevalence (ALP) in a farrowing room. It is expected that the results from this study will
 97 not only provide insights to swine practitioners as to the likely relationship between PP, TLP, and
 98 ALP, but also provide a template/framework for determining the piglet-level prevalence of PRRSV
 99 (and potentially other swine pathogens) that are being monitored using aggregate samples.

100 2. Methodology

101 2.1 PRRSV detection in pig litters using FOF

102 Based on a dataset from ~~Almeida et al.~~ (Almeida, Zhang, Zimmerman, et al., (2021) 199 litters had
 103 all piglets sampled for PRRSV RNA detection by RT-rtPCR (reverse transcription real-time
 104 polymerase chain reaction); also, each litter ($i=1,...,199$) was sampled using FOF. The litters were
 105 sampled from 11 farrowing rooms across six different swine breeding farms ($j=1,...,6$).

106 The effect of the proportion of positive piglets (x) on the detection of a positive litter using FOF
 107 (P^{FOF}) was assessed with a generalized linear mixed model employing a logit link function and a
 108 'residual' Bernoulli distribution. In addition, the linear predictor comprised random effects for farms
 109 according to:

$$110 \text{logit}(P_{ij}^{FOF}) = \alpha + \beta x_{ij} + \varepsilon_{ij} + \gamma_j, \quad (1)$$

111 where α is the intercept of the model. ε_i is the random error assumed $\varepsilon_{ij} \sim N(0, \sigma_\varepsilon)$, and γ_j is the
 112 random effect accounting for the farm-effect in the model, assumed $\gamma_j \sim N(0, \sigma_\gamma)$, where ε_{ij} and γ_j
 113 are all independent. Approximate maximum likelihood inference was based upon Laplacian
 114 integration, as implemented in R (R Core Team, 2018) in routine *glmer* from library *lme4* (Bates
 115 et al., 2015).

116

117 2.2 Stochastic model

Let's consider a random variable: number of positive piglets in the i -th litter (N_i), assuming that each piglet's status (positive/negative) is a Bernoulli trial, with a fixed p probability, thus N_i arises from a binomial process. Consider a room with n litters with different sizes (T_i) drawn from a discrete empirical distribution, and total number of piglets in the room $T = \sum_{i=1}^n T_i$. In a simplistic scenario, the allocation of positive piglets in each litter (N_i) would follow the relative size of the litter in the room. However, given that we are modeling an infectious disease, there might be situations where the total number of positive animals (N) may be "clustered" in a few litters (Carpenter, 2001; Kostoulas et al., 2013).

Accounting for this, the number of positive animals in each i litter (N_i) is calculated as a special case of the multinomial distribution, sampling recursively from binomial distributions using a clustering factor:

$$N_i | N_j (j = 1, \dots, i-1) \sim \min \{ \text{Bin}[(N - \sum_{j=1}^{i-1} N_j, pl_i)], T_i \}, \quad (2)$$

where j stands for the successive allocation of positive animals within each litter. pl is the probability of success in this binomial process. Finally, pl is defined as:

$$pl_i = \frac{T_i}{T - \sum_{j=1}^{i-1} T_j} + \left(1 - \frac{T_i}{T - \sum_{j=1}^{i-1} T_j} \right) \cdot c, \quad c \in [0, 1]. \quad (3)$$

The reader should be aware that c is a clustering factor. Thus when $\lim_{c \rightarrow 1} pl = 1$, the positive piglets will be totally clustered in the smallest number of litters as possible. On the other hand, when $\lim_{c \rightarrow 0} pl = \frac{T_i}{T - \sum_{j=1}^{i-1} T_j}$, piglets will be spread according to the relative size (number of piglets) of each litter regarding the room size.

To obtain the baseline clustering factor c we used the observed distribution of the within litter prevalence (θ_i) reported in Almeida et al. (Almeida, Zhang, Zimmerman, et al., 2021) across seven rooms, each room with n litters. The lost function was the minimization of the mean squared errors of the predicted (eq 2) vs observed distribution of the within litter prevalence $f(c, \theta) = \frac{1}{n} \sum_{i=1}^n (\theta_i - \frac{N_i}{T_i})^2$. The objective function $f(c, \theta)$ can be used to calculate a parameter estimate $\hat{c} = \text{argmin}(f(c, \theta))$. Each room was randomly chosen, 10000 times obtaining the parameters θ_i , N_i , and T_i . For each sampled room, 1000 acquisition points in the parameter space of c were sampled from a uniform distribution $c \sim \text{uniform}(0, 1)$, obtaining a distribution to optimize \hat{c} .

2.2.2 Apparent prevalence at the litter level

147 The simulated proportions of positive piglets per litter obtained from 2.2 were used as input for the
 148 logistic model fit in 2.1, calculating the probability of detection of each simulated litter using FOF
 149 sampling. Now we are interested in modeling a random variable (S) describing the most probable
 150 number of positive litters detected in a routine FOF sampling in a farrowing room. Assuming the
 151 probability of each litter being detected by FOF (P_i^{FOF} see eq. 1) are independent of each other, and
 152 the positive/negative status of a litter $y_i \sim \text{Bernoulli}(P_i^{FOF})$, S equals $\sum_{i=1}^k y_i$. The expected
 153 apparent litter prevalence (ALP) was obtained as S/n . S was generated a total of 2,000 times to
 154 improve the accuracy of the Monte Carlo estimation, and the mean ALP was obtained and stored
 155 for that iterated room.

156 The parameters and distributions used in the simulations are described in table 1. In this simulation,
 157 5000 stochastic iterations were performed, each one representing a different room, propagating the
 158 between litter variability observable in different farrowing rooms.

159 Table 1 – Descriptions of baseline model parameters used to compare the true and apparent liter prevalence
 160 of PRRSV.

Parameter/variable	Distribution/function	Description	Source
p	Fixed= (range of values from 1% to 100%)	Probability of a piglet being positive in a room (prevalence)	Almeida
N	$p \cdot T$	Total number of positive animals in the room	Calculation
T	$\sum_{i=1}^n T_i$	Total number of piglets in the room	Calculation
T_i	empirical $\{(), ()\}^*$	Number of piglets in the i -th litter	Almeida
n	Fixed=56	Number of crates in a room	Authors' opinion
N_i	$\min \{ \text{Bin}[(N - \sum_{j=1}^{i-1} N_j, pI)], T_i \}$	Number of positive piglets in i -th litter	Calculation

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 Your readers need to understand, why to do this

pl_i	$\frac{T_i}{T-\sum_{j=1}^{i-1} T_j} + \left(1 - \frac{T_i}{T-\sum_{j=1}^{i-1} T_j}\right) \cdot c$	Probability of success in this binomial process (i.e., allocation of positive piglets in a litter) for the in i -th litter	Calculation
c	Fixed=0.61	Clustering factor	Optimized based on Almeida

* empirical {(3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 25), (0.0092, 0.0092, 0.0046, 0.0046, 0.0553, 0.0691, 0.0922, 0.1014 0.1982, 0.2074, 0.1244, 0.0783, 0.0415, 0.0046)}

2.3 Sensitivity analysis

To assess the effect of the clustering factor (c) and the room size (n) on the estimated relationship between pig-level-prevalence and litter-level prevalence we selected five values for c (0.05, 0.34, 0.63, 0.83, 1) and five values for n (10, 33, 56, 79, 102) combining them as a factorial design for the sensitivity analysis, totaling 25 different scenarios.

3. Results

3.1 PRRSV detection in pig litters using FOF

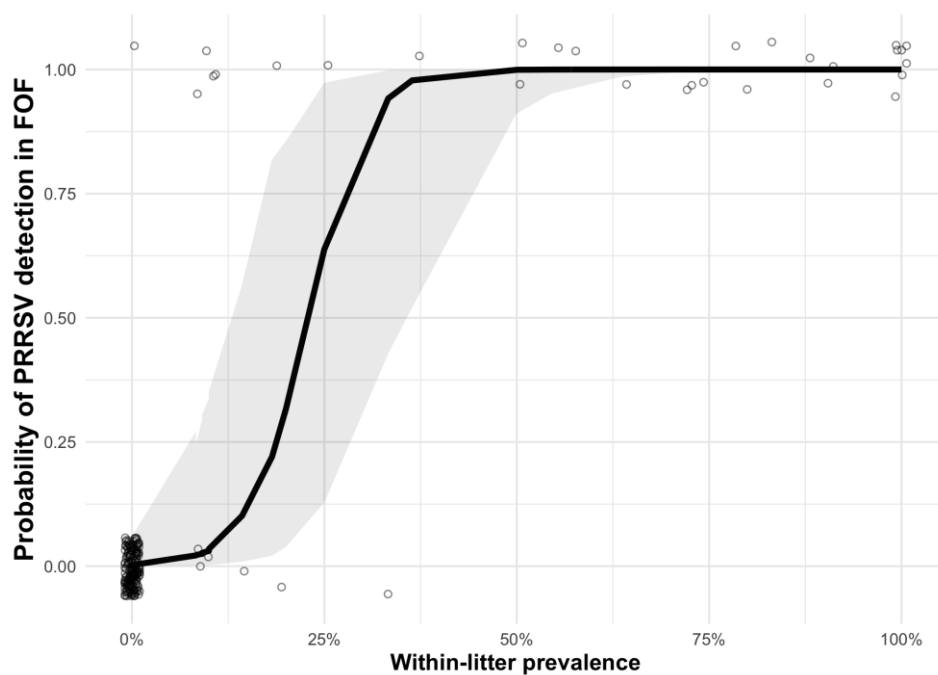


Figure 1: A jitter plot of the Probability of PRRSV detection in FOF by the proportion of positive pigs within litters (within litter prevalence). 95% prediction intervals are represented by the grey region around the regression line.

3.2 Stochastic model

3.2.1 Observed distribution of clustering in sampled farms

The clustering distribution across all sampled rooms had a minimum value of 0.00136, a median of 0.61, a mean of 0.57, and a maximum value of 1. The distributions of the clustering parameter across all sampled rooms are represented in figure 2.

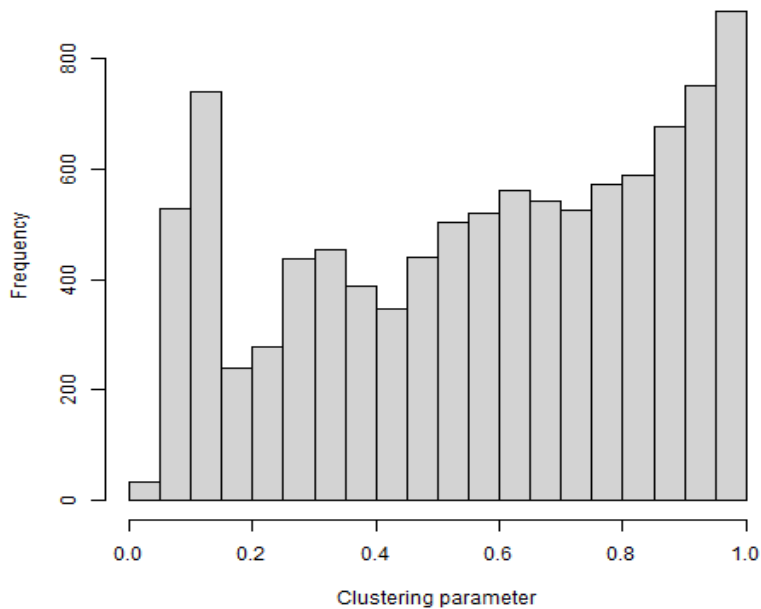


Figure 2: A histogram showing the distribution of the clustering parameter \hat{c} across all sampled rooms from Almeida's study.

3.2.2 The relationship between piglet-level prevalence and litter-level prevalence

Table 2: Relationship between the proportion of positive piglets in a 56-crate farrowing room and the True and Apparent (by FOF) proportion of positive litters assuming a clustering level of 0.61.

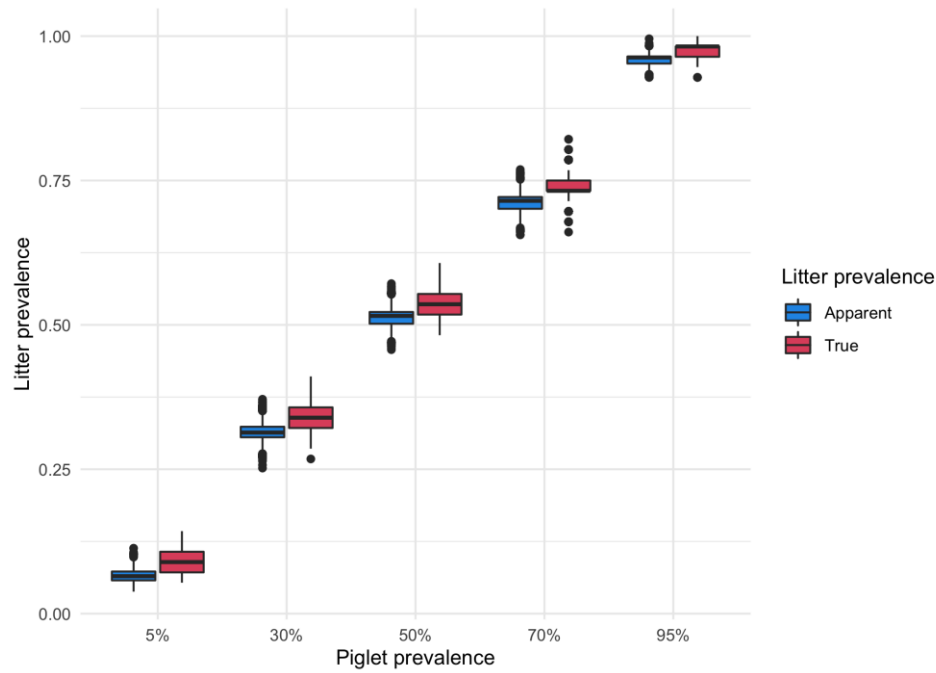
Proportion of PRRSV-positive piglets	True proportion of PRRSV-positive litters (Upper and lower 95% quantiles)	Apparent proportion of PRRSV-positive litters by FOF (Upper and lower 95% quantiles)
(%)	(%)	(%)
1	5.36(1.79,7.14)	2.06(1.07,3.53)
5	8.93(7.14,12.50)	6.48(5.30,8.58)
10	14.29(10.71,17.86)	11.25(9.31,13.92)
15	19.64(16.07,23.21)	16.35(14.47,19.21)
20	23.21(21.43,26.79)	21.60(18.73,24.19)
25	28.57(25.00,32.14)	26.66(23.50,29.31)

30	33.93(30.36,37.50)	31.35(28.77,34.33)
35	39.29(35.71,42.86)	36.16(33.49,39.44)
40	44.64(41.07,48.21)	41.30(38.05,44.71)
45	48.21(44.64,53.57)	46.54(43.10,49.68)
50	53.57(50.00,57.14)	51.56(48.34,54.58)

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195 *Figure 3: Distribution of True- and Apparent litter prevalence in a 56-crate room given different*
196 *piglet-level prevalence scenarios and a clustering factor of 0.63.*

197

198 **3.3 Sensitivity analysis**

199 The sensitivity analysis was done to evaluate the effect of variations in clustering level and room
200 size on the proposed relationship between piglet level prevalence and litter prevalence. As can be

seen from the plots, ALP was relatively more stable to changes in clustering and the number of crates compared to TLP. Generally, TLP and ALP increasingly converged to similar values with increasing clustering and increasing room size. Clustering changes appeared to have a more significant effect on ALP and TLP than changes in the number of crates in the room (Figure 4).

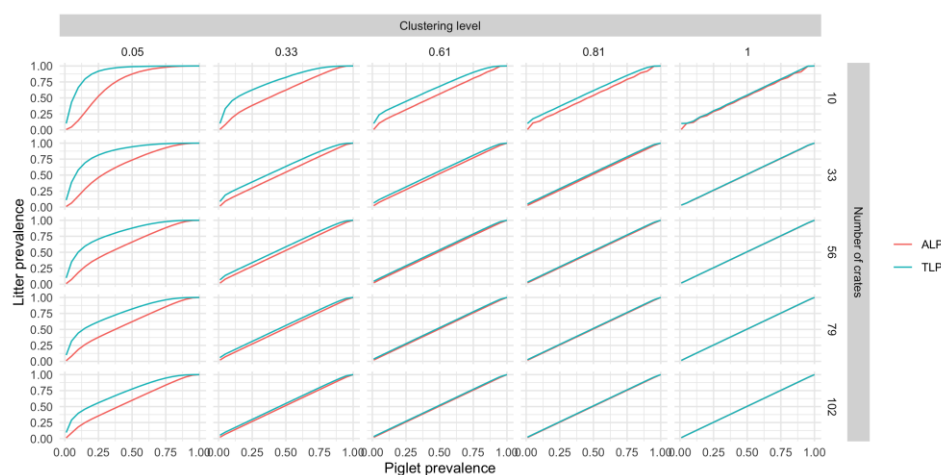


Figure 4: A graphical representation of changes in the relationship between the proportion of PRRSV-positive pigs and the proportion of PRRSV-positive litters (True and Apparent) with changes in clustering of PRRSV within room, and number of litters within rooms.

4. Discussion

The use of mathematical models to describe disease dynamics in swine populations is not new. A few examples include the use of mathematical models to characterize and describe PRRSV transmission dynamics (Amirpour Haredasht et al., 2017; Evans et al., 2010; Nodelijk et al., 2000; Phoo-ngurn et al., 2019; Rotolo et al., 2017; Suksamran et al., 2017) and in the evaluation of PRRSV control strategies (Arruda et al., 2016; Jeong et al., 2014).

218 Earlier studies have described the non-homogenous distribution of PRRSV in pig barns (M. N.
219 Almeida et al., 2021; Rotolo et al., 2017). The non-homogenous aerial distribution of an infectious
220 pathogen however is not limited to PRRSV alone (Carpenter, 2001; Kostoulas et al., 2013), and
221 may be explained by PRRSV being more infectious than it is contagious (Pileri & Mateu, 2016),
222 or by the mere fact that pigs in conventional US barns do not interact randomly with each other and
223 are more likely to have the most/only contact with pigs within the same crate or with their closest
224 neighbors (Murato et al., 2020).

225 Some popular statistical methods used in veterinary epidemiology for detecting and evaluating
226 spatial (areal) clustering include Moran's I , ohno, black-white, Geary's c , and I pop (Carpenter,
227 2001), however, the use of the recursive binomial model in this study offered the authors a
228 simplistic method to not only measure clustering, but to also propagate clustering in simulated
229 data. The use of binomial models to detect and simulate clustering is also not new (Li et al., 2018;
230 Nauta, 2005).

231 The restricted movement of pigs in conventional US swine barns and the non-homogenous
232 distribution of viremic animals have been historically recognized to make conventional sample size
233 assumptions (to detect a disease pathogen) not an exact fit; some previously proposed solutions
234 include replacing simple random sampling with fixed spatial sampling (Rotolo et al., 2017), risk-
235 based sampling (Almeida, Zhang, Zimmerman, et al., 2021)-, or stratified sampling (Almeida,
236 Zhang, Lopez, et al., 2021). This study may well be one more step in trying to adjust conventional
237 sampling schemes to better fit peculiarities with typical swine barns in the United States and with
238 the ecology of PRRSV.

239 Clustering estimates the degree of homogeneity (or more aptly put; heterogeneity) of PRRSV
240 between litters in a farrowing room; it may be overreaching to deterministically model a one-size-
241 fits-all clustering for PRRSV. This is because the spread of PRRSV between litters within a
242 farrowing room would depend on 1) Management practices such as cross-fostering, and vaccination
243 (Mccaw, 2000; Pileri & Mateu, 2016) 2) PRRSV strain (there is evidence of differences in
244 characteristics such as virulence and spread between PRRSV strains) (Cho et al., 2007; Ogno et al.,
245 2019; Pileri & Mateu, 2016) 3) Barn structure (Rotolo et al., 2017) 4) Time since outbreak (Rotolo
246 et al., 2017) 5) Secondary infections which may increase pig susceptibility to PRRSV, encourage
247 huddling or increase production of infectious respiratory fluids.

248 The uncertainty in definitively ascertaining clustering level however does not undermine the
249 importance of these results or pose a challenge to its utilization, on the contrary,
250 considering/estimating clustering adds some precision to the estimated prevalence guiding sample
251 size calculations for disease pathogen surveillance (An example is given in *Table S3*).

252 A critical goal of this study is to estimate the most likely relationship between the pig level
253 prevalence and apparent litter prevalence by FOF, considering the pen-level sensitivity and
254 specificity of this sample type. As observed from Figure 3, ALP is not as sensitive as TLP to
255 variations in clustering parameter; we are therefore confident of the estimates on Table 2. One can
256 also decide the number of crates or litters to randomly sample for FOF to detect PRRSV given an
257 assumed piglet level prevalence. For example, assuming- at least 10% piglet-level prevalence,
258 serum sampling requires that about 30 pigs are sampled to be 95% confident of detecting at least
259 one positive animal (Cannon & Roe, 1982; Holtkamp et al., 2011). From the table, 10% pig-level
260 prevalence corresponds to about 11% ALP or about ~~7~~^{seven} litters in a 56-crate room likely to give
261 a positive FOF test. This number can be used to calculate an appropriate sample size for FOF to
262 detect at least ~~1~~^{one} positive litter; Table 2 is, therefore, useful to the practitioner in tying back what
263 an assumed litter prevalence would be from an assumed piglet-level prevalence.

264 Our approach to calculating ALP implicitly considers the diagnostic performance of FOF sampling;
265 simply put, for a given piglet-level prevalence, the difference between the ALP and TLP is due to
266 the diagnostic performance of FOF (the probability of RT-rtPCR testing of FOF samples to
267 correctly assign PRRSV statuses to each tested litter). This implies that ALP can be used directly
268 to estimate FOF sample size and the only diagnostic performance we may need to consider is that
269 of the RT-rtPCR test kit.

270 Another key application of the proposed tables is to help the swine practitioner estimate his piglet-
271 level prevalence given the results of FOF testing. Given that a representative number of litters were
272 sampled (sample size to estimate prevalence), the proportion of positive FOF results on RT-qPCR
273 tests (apparent litter prevalence by FOF) can be used to deduce the likely proportion of viremic
274 pigs (piglet-level prevalence).

275 The referenced study (Almeida, Zhang, Lopez, et al., 2021) was not designed to answer the question
276 of spatial distribution of viremic piglets within farrowing rooms, as such, in some sampled rooms,

not every litter was sampled; consequently, the observed clustering values for those rooms may be inaccurate. To be able to deduce the number of viremic pigs from FOF positivity rate using the provided tables, it is important that one should have sampled the minimum number of litters needed to estimate prevalence.

5. Conclusion

This study explored the use of mathematical models to characterize the relationship between PPV, TLP, and ALP in a farrowing room, this is a handy surveillance tool for weaning age pigs in typical US swine breeding herds. Another key takeaway from this study is the demonstration of the effect of a clustering parameter on the characterized relationship between the mentioned proportions; like other sampling assumptions, clustering could be considered when estimating sample size. Further similar studies on other aggregate sample types, for other subpopulations and perhaps, for other pathogens will be helpful in guiding practitioners on how they can be up-to-speed with best practice surveillance as sampling methods evolve.

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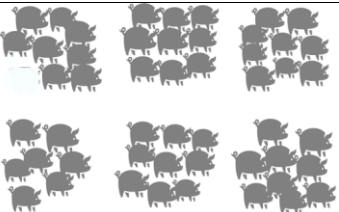
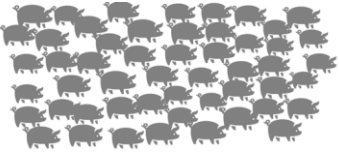
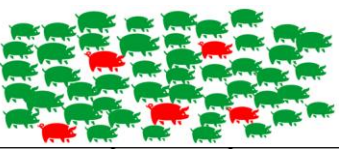
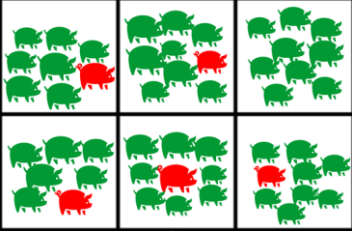
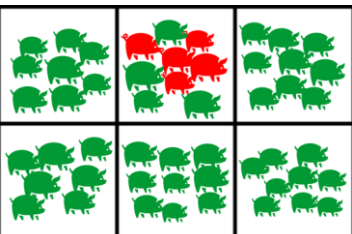
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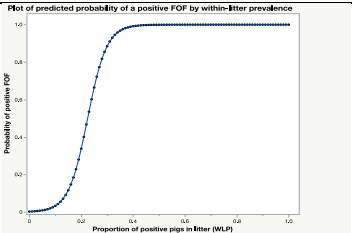
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435 **SUPPLEMENTARY MATERIALS**

436 *Figure S5: A general description of the stochastic model with pictorial illustrations*

	Goal	Example	Example in pictures
1	To create a farrowing room with n litters by generating n	From this empirical distribution, a	

	random numbers from a discrete empirical distribution corresponding to the average litter size we observed from actual field data	possible set of random numbers generated will be 8, 9, 9, 7, 9, 8. These numbers are shown in the picture on the right.	
2.	<p>To create a disease prevalence scenario within this created room.</p> <p>Suppose we wanted a 10% prevalence. We will want 10% of the total pigs in the simulated room to be positive.</p>	<p>When we add all the litters (generated numbers), we have 52 pigs ($8+9+9+7+9+8 = 50$).</p> <p>10% of 50 is 5, meaning five pigs will be PRRSV positive</p>	 
3	<p>To distribute the diseased animals between pens in a manner typical of PRRSV. PRRSV has been repeatedly reported to be heterogeneously distributed (clustered) within a farrowing room. A clustering factor in the recursive binomial model, which could range between 0 (homogenous distribution) and 1 (complete clustering), was used to assign positive pigs to litters.</p> <p>The clustering factor determines the True Litter Prevalence (TLP), defined as the number of litters with at least one positive pig.</p>	<p>The first image shows what would be obtainable if there was no clustering (Clustering = 0). TLP = 5/6</p> <p>The next image shows what is expected in complete clustering (clustering = 1). TLP = 1/6.</p>	 

4	<p>To determine the expected number of positive FOF from this room if all the litters are tested.</p> <p>A predictive model is fitted using data from a previous study (Almeida, Zhang, Lopez, et al., 2021). This model gives the probability of a positive FOF (pFOF) test given the proportion of positive pigs within a litter or within-litter prevalence (WLP).</p>	<p>The graph shows the relationship between pFOF and WLP.</p>  <p>For the clustering = 0, the WLP and pFOF for each litter are calculated.</p> <table border="1" data-bbox="703 703 1070 823"> <tbody> <tr> <td>WLP = 0.125 pFOF = 0.062</td><td>WLP = 0.111 pFOF = 0.043</td><td>WLP = 0 pFOF = 0.002</td></tr> <tr> <td>WLP = 0.143 pFOF = 0.099</td><td>WLP = 0.111 pFOF = 0.043</td><td>WLP = 0.111 pFOF = 0.043</td></tr> </tbody> </table> <p>The expected number of positive FOF for that room is 0.2998 (<1).</p> <p>ALP = <1/6</p>	WLP = 0.125 pFOF = 0.062	WLP = 0.111 pFOF = 0.043	WLP = 0 pFOF = 0.002	WLP = 0.143 pFOF = 0.099	WLP = 0.111 pFOF = 0.043	WLP = 0.111 pFOF = 0.043
WLP = 0.125 pFOF = 0.062	WLP = 0.111 pFOF = 0.043	WLP = 0 pFOF = 0.002						
WLP = 0.143 pFOF = 0.099	WLP = 0.111 pFOF = 0.043	WLP = 0.111 pFOF = 0.043						
5	<p>Repeat steps 1 to 4 (4,999 more times) and obtain the median TLP and ALP thereafter. The 5% prevalence is then matched with the median TLP and median ALP for the chosen clustering level.</p>	<p>For the clustering = 1.</p> <p>The expected number of positive FOF for that room is 1.014 (approximately 1).</p> <p>ALP = 1/6</p> <table border="1" data-bbox="703 1066 1070 1186"> <tbody> <tr> <td>WLP = 0 pFOF = 0.002</td><td>WLP = 0.556 pFOF = 1.000</td><td>WLP = 0 pFOF = 0.002</td></tr> <tr> <td>WLP = 0 pFOF = 0.002</td><td>WLP = 0 pFOF = 0.002</td><td>WLP = 0 pFOF = 0.002</td></tr> </tbody> </table>	WLP = 0 pFOF = 0.002	WLP = 0.556 pFOF = 1.000	WLP = 0 pFOF = 0.002	WLP = 0 pFOF = 0.002	WLP = 0 pFOF = 0.002	WLP = 0 pFOF = 0.002
WLP = 0 pFOF = 0.002	WLP = 0.556 pFOF = 1.000	WLP = 0 pFOF = 0.002						
WLP = 0 pFOF = 0.002	WLP = 0 pFOF = 0.002	WLP = 0 pFOF = 0.002						
6	Then repeat steps 1 to 5 for other prevalence scenarios							

439 Table 3: Relationship between PP, ALP, and TLP at different clustering levels and room sizes

PP (%)	Number of crates (%)	Clustering (%)	ALP (%)	TLP (%)
1.00	10	5.00	0.43	10.00
5.00	10	5.00	3.55	40.00
10.00	10	5.00	14.24	70.00
15.00	10	5.00	27.20	80.00
20.00	10	5.00	40.68	90.00
25.00	10	5.00	52.83	90.00
30.00	10	5.00	63.14	100.00
35.00	10	5.00	71.21	100.00
40.00	10	5.00	78.01	100.00
45.00	10	5.00	83.62	100.00
50.00	10	5.00	87.88	100.00
1.00	10	33.00	0.45	10.00
5.00	10	33.00	8.48	30.00
10.00	10	33.00	19.49	50.00
15.00	10	33.00	27.50	50.00
20.00	10	33.00	32.27	60.00
25.00	10	33.00	38.56	60.00
30.00	10	33.00	42.51	70.00
35.00	10	33.00	48.40	70.00
40.00	10	33.00	52.21	70.00
45.00	10	33.00	58.21	80.00
50.00	10	33.00	61.85	80.00
1.00	10	61.00	0.45	10.00
5.00	10	61.00	10.31	20.00
10.00	10	61.00	17.35	30.00
15.00	10	61.00	20.87	30.00
20.00	10	61.00	27.25	40.00
25.00	10	61.00	30.81	40.00
30.00	10	61.00	37.26	50.00
35.00	10	61.00	40.65	50.00
40.00	10	61.00	46.87	60.00
45.00	10	61.00	50.57	60.00
50.00	10	61.00	56.75	70.00
1.00	10	100.00	0.46	10.00
5.00	10	100.00	10.19	10.00
10.00	10	100.00	10.23	10.00

15.00	10	100.00	20.15	20.00
20.00	10	100.00	20.22	20.00
25.00	10	100.00	30.13	30.00
30.00	10	100.00	30.19	30.00
35.00	10	100.00	40.11	40.00
40.00	10	100.00	40.17	40.00
45.00	10	100.00	50.09	50.00
50.00	10	100.00	50.14	50.00
1.00	56	5.00	0.64	10.71
5.00	56	5.00	7.35	35.71
10.00	56	5.00	18.28	50.00
15.00	56	5.00	27.65	58.93
20.00	56	5.00	34.97	66.07
25.00	56	5.00	41.26	71.43
30.00	56	5.00	46.64	75.00
35.00	56	5.00	51.69	78.57
40.00	56	5.00	56.51	82.14
45.00	56	5.00	61.27	85.71
50.00	56	5.00	66.01	87.50
1.00	56	33.00	1.75	7.14
5.00	56	33.00	7.92	14.29
10.00	56	33.00	13.02	19.64
15.00	56	33.00	18.04	23.21
20.00	56	33.00	23.13	28.57
25.00	56	33.00	27.93	33.93
30.00	56	33.00	32.86	39.29
35.00	56	33.00	37.92	42.86
40.00	56	33.00	42.97	48.21
45.00	56	33.00	47.89	53.57
50.00	56	33.00	52.86	58.93
1.00	56	61.00	2.06	5.36
5.00	56	61.00	6.45	8.93
10.00	56	61.00	11.27	14.29
15.00	56	61.00	16.34	19.64
20.00	56	61.00	21.61	23.21
25.00	56	61.00	26.62	28.57
30.00	56	61.00	31.36	33.93
35.00	56	61.00	36.18	39.29
40.00	56	61.00	41.28	44.64
45.00	56	61.00	46.54	48.21

50.00	56	61.00	51.62	53.57
1.00	56	100.00	2.00	1.79
5.00	56	100.00	5.57	5.36
10.00	56	100.00	10.91	10.71
15.00	56	100.00	16.23	16.07
20.00	56	100.00	20.42	21.43
25.00	56	100.00	25.18	25.00
30.00	56	100.00	30.52	30.36
35.00	56	100.00	35.85	35.71
40.00	56	100.00	41.18	41.07
45.00	56	100.00	45.19	46.43
50.00	56	100.00	50.13	50.00
1.00	102	5.00	0.91	9.80
5.00	102	5.00	8.75	29.41
10.00	102	5.00	18.14	39.22
15.00	102	5.00	24.96	46.08
20.00	102	5.00	30.44	50.98
25.00	102	5.00	35.47	55.88
30.00	102	5.00	40.36	60.78
35.00	102	5.00	45.31	64.71
40.00	102	5.00	50.15	68.63
45.00	102	5.00	54.99	73.53
50.00	102	5.00	59.81	77.45
1.00	102	33.00	2.07	4.90
5.00	102	33.00	6.85	9.80
10.00	102	33.00	11.85	14.71
15.00	102	33.00	16.82	19.61
20.00	102	33.00	21.76	24.51
25.00	102	33.00	26.75	30.39
30.00	102	33.00	31.67	35.29
35.00	102	33.00	36.66	40.20
40.00	102	33.00	41.68	45.10
45.00	102	33.00	46.69	50.00
50.00	102	33.00	51.68	54.90
1.00	102	61.00	1.99	2.94
5.00	102	61.00	6.04	6.86
10.00	102	61.00	10.99	11.76
15.00	102	61.00	15.90	17.65
20.00	102	61.00	20.82	22.55
25.00	102	61.00	25.77	27.45

30.00	102	61.00	30.74	32.35
35.00	102	61.00	35.70	37.25
40.00	102	61.00	40.81	42.16
45.00	102	61.00	45.89	47.06
50.00	102	61.00	50.88	51.96
1.00	102	100.00	1.21	0.98
5.00	102	100.00	5.14	5.88
10.00	102	100.00	10.31	10.78
15.00	102	100.00	15.68	15.69
20.00	102	100.00	20.74	20.59
25.00	102	100.00	25.65	25.49
30.00	102	100.00	30.54	30.39
35.00	102	100.00	35.44	35.29
40.00	102	100.00	40.33	40.20
45.00	102	100.00	45.23	45.10
50.00	102	100.00	50.12	50.00